Bioorganic & Medicinal Chemistry Letters 28 (2018) 1178–1181

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Identification of an aminothiazole series of RORβ modulators

Rémi Patouret, Christelle Doebelin, Ruben D. Garcia-Ordonez, Mi Ra Chang, Claudia Ruiz, Michael D. Cameron, Patrick R. Griffin, Theodore M. Kamenecka *

The Scripps Research Institute, Scripps Florida, Department of Molecular Medicine, 130 Scripps Way #A2A, Jupiter, FL 33458, USA

ARTICLE INFO

Article history:
Received 19 December 2017
Revised 20 February 2018
Accepted 1 March 2018
Available online 3 March 2018

Keywords:
Nuclear receptor
RORβ
Selective ligand
Aminothiazole

ABSTRACT

Crystallography has identified stearic acid, ALRT 1550 and ATRA as ligands that bind RORβ, however, none of these molecules represent good starting points to develop optimized small molecule modulators. Recently, Compound **1** was identified as a potent dual RORβ and RORγ inverse agonist with no activity towards RORα (Fig. 1). To our knowledge, this is one of only two small molecule RORβ inverse agonists identified in the primary literature from a tractable chemical series and represents an ideal starting point from which to design RORβ-selective modulators. Herein we describe our SAR optimization efforts that led to a series of potent neutral antagonists of RORβ.

The nuclear receptor (NR) superfamily of ligand-regulated transcription factors has proven to be a rich source of targets for the development of therapeutics for a wide range of human diseases. Retinoic acid receptor-related orphan receptors (RORs) are transcription factors that belong to the steroid hormone nuclear receptor super family. Several nuclear receptors are characterized as orphan receptors because the ligands for these receptors are still unknown or controversial. The NR1F subfamily, known as the retinoic acid receptor-related orphan receptors (RORα [NR1F1], RORβ [NR1F2] and RORγ [NR1F3]), regulate several physiological processes, including the circadian rhythm, glucose and lipid metabolism, and immune functions. RORα is expressed in the liver, skeletal muscle, skin lungs, adipose tissue, kidney, thymus and brain.[1,2] RORγ is most highly expressed in the thymus, but significant expression is also found in the liver, skeletal muscle, adipose tissue and kidney.[3] RORβ has a more restricted expression pattern and is found in regions of the central nervous system (CNS) that are involved in processing of sensory information and components of the mammalian timing system (circadian clock), including the suprachiasmatic nuclei (SCN), retina, pineal gland and bone.[4,5] RORβ−/− mice show defects in circadian rhythmicity.[4] Aberrant circadian rhythms are associated with numerous ailments in humans including bipolar disorder, schizophrenia, major depressive disorder and seasonal affective disorder.[6–12] RORβ is expressed in the retina and genetic deletion of RORβ results in retina degeneration, implicating its role in vision development. RORβ null mice suffer from retinal degeneration, and are born blind.[13] Most recently, it was discovered that RORβ plays a role in osteogenesis by impacting Runx2 expression.[14]

Levels of RORβ inversely correlate with osteogenic potential, suggesting that suppression of RORβ may drive osteoblast mineralization. While RORα−/− mice displayed bone abnormalities, the bone phenotype in RORβ−/− mice has not been characterized. RORβ and a subset of RORβ-regulated genes were found to be overexpressed in bone biopsies from post-menopausal women, suggesting a role for RORβ in human age-related bone loss.[15]

While the *in vivo* functions of RORα and RORγ have been widely explored over the past decade, the exact roles for RORβ still remain elusive, in part due to lack of specific molecular probes that would enable investigation of its function. Previously it was shown that all trans retinoic acid (ATRA) and a synthetic analog (ALRT 1550) bind to RORβ and are functional inverse agonists, but these ligands lack potency and bind other nuclear receptors including the RXRs and RARs.[16,17] Stearic acid was also shown to bind RORβ during expression and purification, but does not activate the receptor.[18,19]

Our goal was to identify, characterize and develop potent RORβ selective modulators with sufficient ADME properties to facilitate their use as *in vivo* probes. Recently, Phenex Pharmaceuticals identified a potent dual RORβ and RORγ inverse agonist with no activity toward RORα (Compound **1**, Fig. 1).[20] To our knowledge, this is one of only two small molecule RORβ inverse agonists identified in the primary literature from a tractable chemical series and represents an ideal starting point from which to design RORβ selective modulators.[21]

Compound **1** can be dissected into three fragments of equal size and complexity (Fig. 2). We were curious to see which portions of

* Corresponding author.
E-mail address: kameneck@scripps.edu (T.M. Kamenecka).

https://doi.org/10.1016/j.bmcl.2018.03.001
0960-894X/

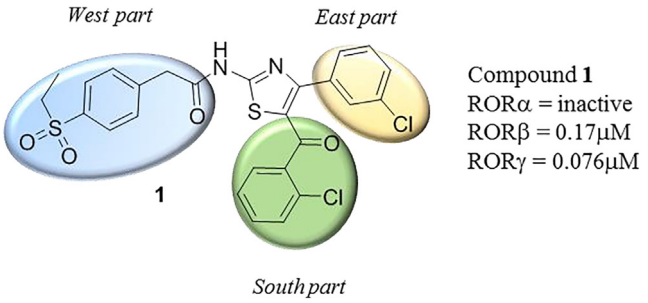


Fig. 1. Lead dual RORβ/RORγ antagonist.

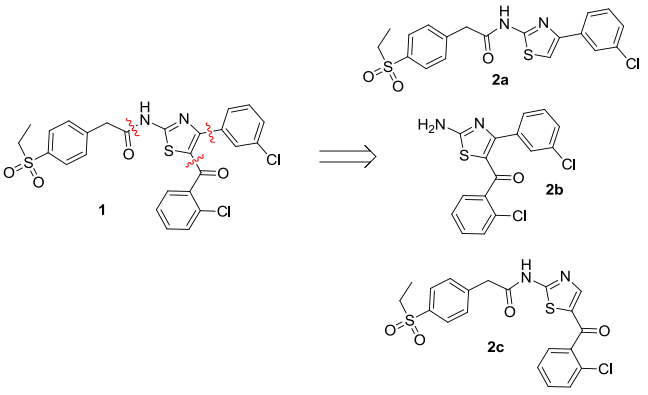


Fig. 2. Truncations of compound **1**.

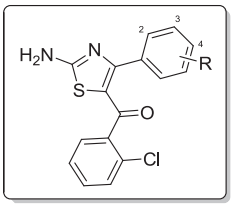
the molecule were absolutely required for activity, so we synthesized three compounds each lacking one portion of the parent molecule (Fig. 2, **2a**-c). The binding potency of the analogs was determined in a scintillation proximity assay (SPA) using ^{3}H T0901317 and recombinant human RORβ and RORγ LBDs.[22] This assay measures the affinity of the compounds for RORβ vs RORγ and is tabulated in Table 1.

Removal of the benzoyl portion led to **2a** and a significant reduction in activity against RORβ as did removal of the C-4 aryl group (**2c**, Table 1). Much to our surprise, truncation of the west part afforded aminothiazole **2b**, which was selective for RORβ vs RORγ. This was unexpected as the ligand binding pocket in RORβ is actually larger than it is in RORγ (766^{3}Å versus 705^{3}Å, respectfully).[23] Despite a ten-fold drop in potency vs compound **1**, this was the first RORβ modulator identified devoid of RORγ activity, and served as the basis for our SAR campaign described herein.

We decided to continue our SAR exploration of **2b** and begin with modifications to the East part of the structure (Table 2). Synthesis of these aminothiazole analogs was straightforward following chemistry as described in the primary literature and is shown in Scheme 1.[24]

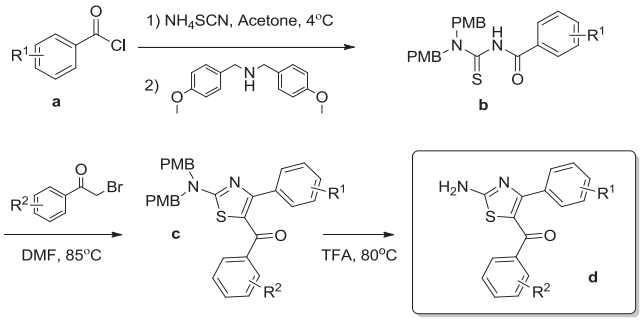
The chlorine substitution was best tolerated at the 3-position in the phenyl ring, but was not tolerated at the 2- and 4-positions (**2b**, **3a**, **3b**, respectively, Table 2). A scan of the ortho-(**3c**), meta-(**3d**), and *para*-positions (**3e**) of the phenyl ring with a bromine atom showed a similar trend, with the 3-position optimal for potency. Incorporation of additional substituents at the 3-position revealed a striking preference for hydrophobic groups (**3g**, **3 k**), whereas polar residues led to loss of affinity (**3f**, **3 h**, **3i**, **3j**). Given the bias

Table 2
East part SAR – Substituted aryl rings.



Cmpd	R	RORβ IC_{50} μM[a]	RORγ IC_{50} μM[b]
2b	3-Cl	0.24 ± 0.05	>40
3a	2-Cl	(50%)	n.t.
3b	4-Cl	(30%)	n.t.
3c	2-Br	(15%)	(20%)
3d	3-Br	0.41 ± 0.13	(15%)
3e	4-Br	(25%)	n.t.
3f	3-OMe	(10%)	n.t.
3g	3-CF_3	0.61 ± 0.06	>40
3h	3-NO_2	(15%)	n.t.
3i	3-CN	(5%)	n.t.
3j	3-SO_2Me	(30%)	n.t.
3k	3-Ph	1.4 ± 0.55	nt
3L	3-Cl, 5-CF_3	0.14 ± 0.018	(10%)
3m	3-Br, 5-CF_3	0.063 ± 0.01	>40
3n	3,5-CF_3	0.31 ± 0.074	13.4
3o	3,5-Br	(60%)	n.t.
3p	3,5-Dimethyl	(55%)	n.t.
3q	3-CF_3, 4-Br	0.29 ± 0.076	(10%)
3r	3-CF_3, 4-Cl	0.12 ± 0.048	>40

[a] Displacement of [^{3}H]-T09 from human RORβ LBD. Values are the mean ± SEM of at least three replicates. IC_{50} or displacement of [^{3}H]-T09 at 1 μM). [b] Displacement of [^{3}H]-T09 from human RORγ LBD. Values are the mean ± SEM of at least three replicates. IC_{50} or displacement of [^{3}H]-T09 at 1 μM); n.t. = not tested.



Scheme 1. Synthesis of aminothiazole analogs.

Table 1
Binding affinity of truncated analogs.

Compound	RORβ%inhib[a]	RORγ%inhib[a]	RORβ IC_{50} μM[b]	RORγμ IC_{50} μM[c]
1	80	85	0.059 ± 0.02	0.013 ± 0.001
2a	30	n.t.		
2b	50	2	0.24 ± 0.05	>40
2c	35	n.t.		

[a] Percent displacement of [^{3}H]-T09 at 1 μM. [b] Displacement of [^{3}H]-T09 from human RORβ LBD. Values are the mean ± SEM of at least three replicates. [c] Displacement of [^{3}H]-T09 from human RORγ LBD. Values are the mean ± SEM of at least three replicates; n.t: not tested.